Characterization and evaluation of onion (*Allium cepa* L.) germplasm at Leh, trans-Himalaya, India

SYNOPSIS

Submitted in fulfillment of the requirements for the degree of

Doctor of Philosophy in

Biotechnology

By

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INTRODUCTION

Farming at high altitude is a challenging task owing to the geography and extreme climatic conditions. Onion is an important food crop grown and consumed almost all over the world and is cultivated in atleast 170 countries. Onion (*Allium cepa*var. cepa L.) that belongs to the family Alliaceae is the second most valuable vegetable crops grown throughout the world after potato rightly called the "queen of the kitchen" (Selvaraj, 1976).It is being used as a condiment in most cuisines and one of the oldest bulb crops known to humans, and is believed to have originated in Central Asia. Onion is consumed universally in small quantities or several times daily in some homes, primarily as a seasoning for flavouring of dishes, soup, sauces, and sandwiches. Onion is often used as a part of traditional medicines, and has been shown to be effective in preventing heart diseases and other diseases (Augusti, 1990). Its medicinal and therapeutic effects include antiplatelet aggregation, blood sugar lowering, and fibrinolytic effects.

However, owing to the challenges at high altitude, it is difficult to produce onion having good quality, good shelf life, and as per the demand of Army deployed in Ladakh and local populace. Though onion is an indispensible ingredient in numerous Indian recipes and is also a 'protective supplementary food", it is, at times, in short supply for the deployed Armed forces and also to civilians in the region of Leh-Ladakh (J&K), owing to the lack of knowledge of appropriate production techniques for onion in the harsh climatic conditions of the region to local farmers. Ladakh is actually a high altitude cold arid zone of India, which is situated in the high mountains of the north-western Himalayas. The trans-Himalayan zone, which constitutes the Indian cold arid zone, is confined to Ladakh (J&K), Lahual&Spiti (H.P.) and a small land mass in Uttaranchal (Niti and Mana Garhwal). Ladakh makes up 87.4 percent of the total cold arid zone of India and it is geographically located between 32°15′ to 36° north latitude and 75°15′ to 80°15′ east longitude (SKUAST-Leh, 1998).

There is a need to find suitable genotypes for commercial production at high altitudes which have high yielding, mineral content and better storability. In the present work, the variability, interrelationship, and divergence pattern of twenty six accessions of onion at high altitude were studied based on quantitative and qualitative traits. Multivariate analysis was used to classify the twenty six onion accessions. The genotypes were divided in different clusters and their characterstics viz., plant height (cm), number of leaves per plant, leaf length (cm), leaf

width (cm), chlorophyll content (cci), neck thickness (cm), total soluble solids (%), dry matter (%), polar diameter (cm), equatorial diameter (cm), bulb shape index, average bulb weight (g) and bulb yield (kg/plot) and four qualitative traits viz., colour of foliage, degree of leaf waxiness, uniformity of bulb shape, colour of bulb skin were studied.

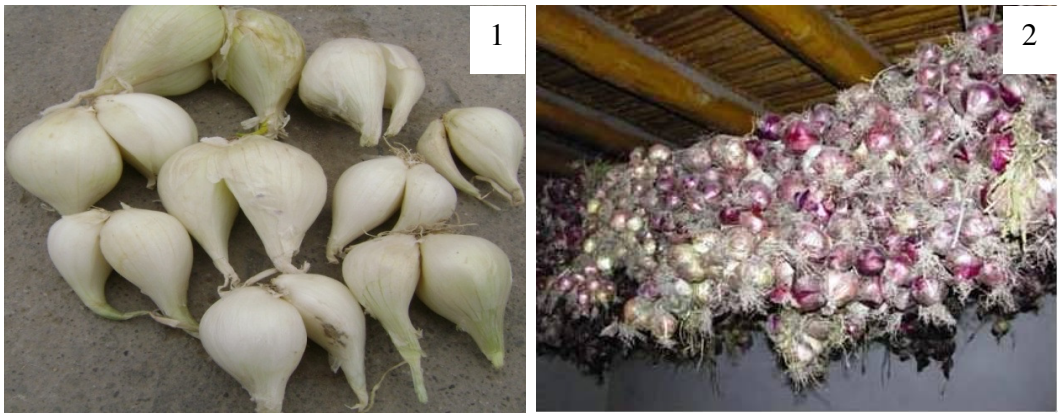

Fig. 1: 1-local cultivar; 2-traditional method of onion storage.

We further investigated the comparative performance of long day onion germplasm using 'Sets and Seedlings' as planting material to study the effect of two planting materials (set and seedling) and genotypes on bulb yield, its quality and vegetative growth. The results indicated that all the genotypes under set planting performed better than seedling ones. Based on our study, three genotypes could be recommended for cultivation in Leh and further studies should be carried out on mineral content and storage potential of the genotypes. These genotypes can be used as parents for crop improvement.

The study was carried out to evaluate the variability of mineral accumulation of in onion genotypes revealed by multivariate analysis for use in further breeding programs to achieve improved cultivars, targeting high nutrient concentrations. A total of 10 mineral elements in10 onion genotypes were investigated, and considerable variation was observed in most of the concentrations. According to the significant concentrations of various minerals found in different clusters, parents could be selected for hybridization or cultivar development programs. Onion genotypes were classifies into three groups and outcomes of the experiments revealed that the first three principal components were highly effective in classifying the examined genotypes on the basis of mineral accumulations. Significant differences exhibited in mineral concentrations

among the examined onion genotypes and the result could help in the selection of those genotypes with higher concentration of elements.

Onion bulbs of various long-day genotypes were stored for different storage durations in controlled environments various parameters were recorded throughout the storage period. For the storage periods, similar patterns of increase and decrease in all the observed traits were shown by all the genotypes. On the other hand, at the genotypic level, a significant variation was observed in their storage potential. From the study, some onion genotypes were characterized as having good storage potential; they could be stored over winter at high altitudes and the supply could be maintained over winter. Meanwhile, this could help extend the onion bulb availability period in temperate regions.

This study resulted in the selection of the genotypes which have high yield, good quality in terms of mineral content and shelf life of onion by which further cultivar improvement programme would be initiated specially for the improvement of local cultivar. Moreover, these genotypes might be used for the development of potential hybrids in onion. It could be recommended as cultivar for the production of onion bulb. This would fill the gap of 65 percent of onion supply to the army and strengthen the socioeconomic condition of civic of Ladakh.

The present investigation was carried out to elucidate the technology for specific varieties along with different crop-management practices for the rapid enhancement of onion production in the agro-climatic conditions of Leh-Ladakh. Therefore, the objective of this study is to understand

1. Morphological variations and relationship among onion germplasms for quantitative and qualitative traits in the trans-Himalayan Ladakh region of India.
2. Comparative performance of onion genotypes using ‘Sets and Seedlings’ as planting material at Leh cold desert.
3. Evaluation of different onion genotypes for mineral content as measured by ICP-OES at agroclimatic condition of Leh, trans-Himalaya, India
4. Evaluation of onion genotypes for post-harvest keeping quality of the bulb in agro-climatic conditions of Ladakh.

1. Morphological variations and relationship among onion germplasm for quantitative and qualitative traits at trans-Himalaya Ladakh, India

Materials and Methods

The experiment was conducted from May to October in 2013 and 2014 in the Vegetable Research Unit at the Defence Institute of High Altitude Research, Leh-Ladakh, (altitude 3500 m above MSL, latitude 34°8'16.119" N, longitude 77°34'19.2216"E). The soil texture of the experimental site was silty loam with pH 7.1±0.2. Organic carbon and organic matter content were 1.2.±0.4% and 4.2±0.5%, respectively.

The experimental material consisted of 26 accessions including a local cultivar (table 1) of onion having a broad spectrum of variation which was obtained from NBPGR New Delhi.

Table 1. List of onion genotypes bring from NBPGR, New Delhi, India

S.no.	Genotypes	Source
1	IC-0279582	National Bureau of Plant Genetic Resources
2	IC-0035182	National Bureau of Plant Genetic Resources
3	IC-0035851	National Bureau of Plant Genetic Resources
4	IC-0035856	National Bureau of Plant Genetic Resources
5	IC-0035891	National Bureau of Plant Genetic Resources
6	IC-0035940	National Bureau of Plant Genetic Resources
7	IC-0035992	National Bureau of Plant Genetic Resources
8	IC-0038823	National Bureau of Plant Genetic Resources
9	IC-0047928	National Bureau of Plant Genetic Resources
10	IC-0047940	National Bureau of Plant Genetic Resources
11	IC-0279180	National Bureau of Plant Genetic Resources
12	IC-310990	National Bureau of Plant Genetic Resources
13	IC-0338582	National Bureau of Plant Genetic Resources
14	IC-0356095	National Bureau of Plant Genetic Resources
15	IC-0369521	National Bureau of Plant Genetic Resources
16	IC-0374706	National Bureau of Plant Genetic Resources
17	IC-0374724	National Bureau of Plant Genetic Resources
18	IC-0391544	National Bureau of Plant Genetic Resources
19	IC-0392640	National Bureau of Plant Genetic Resources
20	IC-0392647	National Bureau of Plant Genetic Resources
21	IC-0440229	National Bureau of Plant Genetic Resources
22	IC-0512324	National Bureau of Plant Genetic Resources
23	IC-0524211	National Bureau of Plant Genetic Resources
24	IC-0588677	National Bureau of Plant Genetic Resources
25	IC-0039039	National Bureau of Plant Genetic Resources

26	Local cultivar	Seeds of Local Cultivar were collected from Local Market of Leh-Ladakh.

The experiment was laid down in randomized complete block design (RCBD) with five replications and one local check; all the recommended agronomic practices and plant protection measures were followed. The observations were recorded from ten randomly selected plants from each accession in each replication for thirteen quantitative characters. The traits were observed as per International Plant Genetic Resources Institute (IPGRI) plant descriptors for *Allium species.* Mean values of all observations were calculated for quantitative traits and subjected to analysis carried out by SPSS 21 statistical analysis software.

Genetic diversity was studied using the Mahalanobis (1936) generalized distance (D^2) extended by Rao (1952). Clustering of genotypes was done according to Tocher's Method. The average intra-cluster distance was calculated by the following formula as suggested by Singh and Choudhary (1985). Trait variability analysis was performed by the principal component analysis (PCA) method, with the number of principal components being chosen based on the screen test (Kovacic, 1994). Agglomerative Hierarchical cluster analysis was used to determine differences and similarities among the genotypes, and the distance measure used was Gower General Similarity Coefficient as the parameter that best reflects the differences existing among the genotypes (Kendall, 1980).

Results:

Based on the D^2 value estimates of genetic divergence the 26 onion genotypes were grouped into four distinct clusters (table 2). A wide range of diversity was observed in the experimental material for the majority of the characters studied. Cluster IV consisted of a maximum of 12 genotypes, Cluster II consisted of 10 genotypes, cluster I consisted of 2 genotypes and cluster II also consisted of 2 genotypes.

Table 2.Distribution of twenty six genotypes of onion in different clusters

Sr.no.	Cluster	Number of genotypes	Genotypes
1	I	2	IC-0392640 , IC-0440229
2	II	10	IC-0035856, IC-0338582, IC-0356095, IC-0369521, IC-0588677, IC-0374724, IC-0392647, IC-0038823, Local Cultivar
3	III	2	IC-0512324, IC-0524211
4	IV	12	IC-0035851, IC-0035940, IC-0374706, IC-310990, IC-0279582, IC-0035182, IC-0035891, IC-0035992, IC-0047928, IC-0047940, IC-0279180, IC-0391544

Cluster II had the highest intra-cluster distance (2.778) followed by cluster IV (2.345), cluster I (2.191) and cluster III (2.064) table 5. The intra cluster distance in all the four clusters was essentially low, indicating that genotypes within the same cluster were closely related. While considering the inter-cluster distance (table 3), maximum inter-cluster distance (6.129) was noticed between I and IV followed by (5.675) between II and IV and (4.933) between I and III. Minimum distance (2.542) was noticed between I and II (table 3).

Table 3.Average intra- (bold face) and inter-cluster distance (D^2) of 26 onion genotypes

Sr. no.	Clusters	I	II	III	IV
1	I	**2.191**			
2	II	2.542	**2.778**		
3	III	4.933	3.649	**2.064**	
4	IV	6.129	5.675	4.645	**2.345**

According to Ghaderi et al. (1984), increasing parental distance implies a great number of contrasting alleles at the desired loci, and to the extent that these loci recombine in the F2 and F3 generation following a cross of distantly related parents, the greater will be the opportunities for the effective selection of yield factors. Thus, the crossing of genotypes from these clusters with other clusters may produce higher amounts of heterotic expression in the first filial generations (F1's) and a wide range of variability in subsequent segregating (F2) populations. Similar results were found by Akter et al., 2015.

Fig. 2 Potential genotypes from 26 accessions of onion.

Based on cluster means (table 4); the important cluster was I for number of leaves (12.90)and neck thickness (1.02 cm), Cluster II for leaf width (3.02 cm), double/deformed bulb(27.90 %), polar diameter (5.93 cm), equatorial diameter (6.43 cm), average bulb weight (92.55 g) and bulb yield kg/plot (18.52 kg/plot), Cluster III for days to maturity (133.60), TSS (12.48 %) and dry matter (14.18 %) and Cluster IV for plant height (55.43 cm), leaf length (46.43 cm) and chlorophyll content (44.34 cci).

Table 4. Mean values for four clusters based on fifteen morphological traits

Sr. no.	Traits	Clusters			
		I	II	III	IV
1	Plant Height (cm)	50.64	38.15	41.83	55.43
2	Number of Leaves	12.90	8.43	12.87	12.86
3	Leaf Length (Cm)	43.42	32.97	38.35	46.33
4	Leaf Width (Cm)	1.16	3.02	1.09	1.20
5	Chlorophyll content	39.14	42.32	37.88	44.34
6	Days to maturity	134.26	141.53	133.60	135.95
7	Double/Deformed Bulb (%)	53.38	27.90	38.03	70.86
8	Neck Thickness(cm)	1.02	1.04	1.74	1.53
9	TSS %	7.90	9.40	12.48	8.58
10	Dry matter %	9.83	11.37	14.18	10.32
11	Polar adiameter (cm)	3.13	5.93	3.60	3.36
12	Equatorial Diameter (Cm)	3.06	6.43	3.43	2.89
13	Bulb shape index	1.03	0.92	1.07	1.17

14	Average bulb wt. (g)	42.94	92.55	70.50	36.48
15	Bulb yield kg/ plot	8.57	18.52	14.14	7.36

On the basis of results; it can be concluded that genotypes that belong to cluster I are suitable for number of leaves and neck thickness. The suitable genotypes for leaf width, double/deformed bulb, polar diameter, equatorial diameter, average bulb weight and bulb yield kg/plot belonged to cluster II. Genotypes which belonged to cluster III are suitable for days to maturity, TSS and dry matter content and cluster IV which was suitable for plant height, leaf length and chlorophyll content could be selected as a parent for the hybridization program. Crosses involving parents belonging to more divergent clusters would be expected to manifest maximum heterosis and wide variability in genetic architecture (Singh et al., 1987; Akter et al., 2015.).

2. **Comparative performance of onion genotypes using 'Sets and Seedlings' as planting material at Leh cold desert.**

Materials and methods

The experimental material consisted of 10 genotypes of onion viz. Red Cereole, Katarina Red 3, Katarina Red 7, Supreme, Cyrus, Lock Roy, Legend, Wall Brown, Brown Spanish and a Local Cultivar having broad spectrum of variation that was received from BejoSheetal Seeds. The experiment was laid down in Randomized block design (RBD) with five replications. The plot size for each treatment was 2m×1m.

Fig. 3: 1,2,3- planting through sets, 4-planting through seedling

Sets and seedlings of all the genotypes selected were of uniform size based on visual observations and transplanted on flat bed system. The plating materials of particular density were transplanted on flat beds in their respective plots with constant 20 cm intra-row distance, while the in-row spacing was 10 cm. Well rotten farmyard manure (FYM) at the rate of 20 t ha^{-1} was incorporated to the soil 30 days before transplanting the seedlings. The recommended dose of NPK fertilizes was applied at a ratio of 80- 60-60 kg ha^{-1}. All the Phosphorous, Potash and half dose of Nitrogen was applied during transplanting the seedlings while the remaining half dose of Nitrogen was applied after 30 days. All the cultural and management practices viz. irrigation, weeding, hoeing and sprays for insect pests and disease control etc. were carried out uniformly for all treatments. Observations were recorded from 10 randomly selected plants from each

genotype in each replication for fourteen traits viz., Plant survival (%), Plant Height (cm), Number of leaves per plant, Leaf Area (cm^2), Chlorophyll Content (CCI), Neck Thickness (cm), Total Soluble Solids (%), Dry Matter (%), Polar Diameter (cm), Equatorial Diameter (cm), Bulb Shape Index, Average Bulb Weight (g) and Bulb Yield (Kg/Plot). Data recorded over 2 years were pooled due to a non-significant heterogeneity test (Azmat*et al.*, 3). Means of all observations were calculated for quantitative traits and subjected to SPSS 21 statistical software. Two way ANOVA with 2-sided Tukey's HSD at $p \leq 0.05$ was carried out.

Results:

There was significant effect of both the two factors under study(Planting material and genotypes) on all the studied parameters. Wall Brown showed highest plant survival (98.15 %), chlorophyll content (39.88 cci), days to maturity (137.60), Polar diameter (6.84 cm), Equatorial diameter (9.94 cm), average bulb weight (185.04 g) and bulb yield (36.32 kg/plot) when grown through set among all the genotypes. On the other hand, highest plant height (42.14 cm) and number of leaves (16.02), leaf area (157.02), were obtained in the genotype Brown Spanish. It was observed that highest TSS (13.90 %) and dry matter (15.50 %) were observed in the genotype Red Cereole. However, lowest double/deformed bulbs (0.74 %) and neck thickness (0.61cm) was observed in the genotypes Lock Roy and Red Cereole, respectively.

Table 5. Effect of set and seedling on plant survival, plant height and number of leaves of onion

	Plant survival (%)		Plant height (cm)		Number of leaves	
Genotypes	Seedling	Set	Seedling	Set	Seedling	Set
Red Cereole	57.70±4.35^{abc}	97.55±1.92^{d}	38.46±2.76^{cd}	41.12±2.64^{bc}	8.52±1.42^{cd}	9.30±1.16^{b}
Katarina Red 3	56.25±7.65^{abc}	93.80±3.72^{d}	29.75±3.50^{ab}	31.04±3.62^{a}	4.92±0.88^{a}	5.15±0.53^{a}
Katarina Red 7	61.45±3.85^{cd}	97.60±1.19^{b}	33.58±3.03^{abc}	35.55±2.57^{ab}	6.24±0.66a^{bc}	6.28±0.94^{b}
Supreme	61.70±2.39^{cd}	97.65±1.93^{b}	37.29±2.01^{bcd}	38.95±1.10^{ab}	9.25±0.78^{de}	9.56±0.65^{bc}
Cyrus	52.30±3.91^{b}	92.25±2.63^{a}	36.65±4.38^{abcd}	39.61±4.71^{ab}	8.22±1.62b^{cd}	9.06±0.78^{b}
Lock Roy	60.75±3.54^{cd}	98.00±1.13^{b}	32.18±1.93^{abc}	34.44±1.26^{ab}	7.06±0.71^{abcd}	7.39±0.67^{ab}
Legend	54.60±2.97^{c}	97.30±2.68^{b}	32.18±1.34^{abc}	34.28±1.02^{ab}	8.28±2.06^{cd}	9.35±2.55^{c}
Wall Brown	64.20±1.92^{d}	98.15±1.27^{b}	33.27±4.31^{abc}	35.39±5.28^{ab}	5.84±0.59^{ab}	6.23±0.90^{a}
Brown Spanish	50.90±3.31^{b}	94.80±2.27^{a}	42.74±5.53^{d}	48.28±8.55^{c}	13.87±1.06^{f}	16.02±0.98^{d}
Local Cultivar	61.35±3.73^{cd}	94.45±3.21^{ab}	28.93±5.21^{a}	34.11±3.40^{ab}	11.02±0.85^{e}	11.74±1.00^{c}

Mean	58.11±5.63	96.16±2.94	34.50±5.25	37.28±5.97	8.32±2.74	9.01±3.20

Value (Mean± SE) bearing common superscript (abcd) within column does not vary significant.

Table 6. Effect of set and seedling on leaf area, chlorophyll contentand days to maturity of onion

	Leaf Area (cm^2)		Chlorophyll content(cci)		Days to maturity	
Genotypes	Seedling	Set	Seedling	Set	Seedling	Set
Red cereole	47.94±9.03^{a}	65.48±9.39^{b}	39.63±2.11^{ab}	39.79±0.54^{bc}	169.10±1.08^{a}	131.50±1.06^{b}
Katarina Red 3	23.33±7.49^{a}	27.10±8.35^{a}	33.24±1.41^{a}	33.77±0.89^{a}	169.20±1.92^{a}	131.90±1.08^{b}
Katarina Red 7	41.91±7.80^{b}	49.61±7.71^{cd}	32.43±0.93^{a}	32.24±0.98^{a}	171.40±1.34^{b}	133.10±1.29^{a}
Supreme	46.79±6.53^{ab}	59.32±6.21^{ab}	35.24±2.25^{ab}	39.30±3.96^{bc}	173.80±1.10^{b}	134.40±1.02^{a}
Cyrus	52.94±8.79^{ab}	61.26±7.28^{ab}	37.80±5.82^{ab}	43.27±2.69^{c}	177.20±1.30^{c}	135.75±1.46^{ab}
Lock Roy	35.09±8.43^{ab}	40.60±11.11^{ab}	34.31±2.30^{a}	36.70±3.37^{ab}	172.40±0.82^{c}	135.10±0.82^{b}
Legend	38.86±10.98^{ab}	43.93±7.00^{abc}	36.60±2.27^{ab}	37.75±2.40^{ab}	177.00±1.00^{c}	135.70±11.39^{b}
Wall Brown	56.51±30.71^{b}	65.53±10.04^{b}	39.86±2.04^{b}	39.88±2.35^{bc}	178.10±0.55^{c}	137.60±0.82^{ab}
Brown Spanish	129.89±17.17^{c}	157.07±23.57^{d}	36.39±1.88^{a}	40.24±4.27^{bc}	176.70±0.57^{c}	135.70±0.82^{ab}
Local Cultivar	35.04±5.09^{ab}	42.80±5.15^{ab}	34.84±1.52^{ab}	35.76±1.50^{ab}	172.80±1.35^{b}	132.40±0.42^{a}
Mean	50.83±31.02	61.22±37.68	35.66±3.13	37.82±3.93	173.77±3.37	134.96±4.64

Value (Mean± SE) bearing common superscript (abcd) within column does not vary significant.

Table 7. Effect of set and seedling on double/deform bulb, neckthickness and TSS of onion bulb

	Double/deform bulb (%)		Neck thickness(cm)		Total soluble solids (%)	
Genotypes			Seedling	Set	Seedling	Set
Red cereole	4.77±0.44^{b}	5.32±0.84^{b}	0.64±0.08^{a}	0.61±0.06^{a}	13.14±0.85^{d}	13.90±0.58^{d}
Katarina Red 3	4.41±0.49^{b}	4.88±0.30^{b}	1.00±0.07^{a}	1.01±0.08^{b}	12.53±0.93^{cd}	13.69±0.42^{d}
Katarina Red 7	1.96±1.27^{a}	2.26±0.91^{a}	0.77±0.06^{a}	0.71±0.04^{ab}	11.92±1.31^{cd}	13.28±1.18^{d}
Supreme	1.13±0.72^{a}	1.26±0.56^{a}	0.81±0.14^{a}	0.72±0.14^{ab}	8.29±0.37^{b}	9.19±0.41^{b}
Cyrus	0.86±0.30^{a}	0.84±0.32^{a}	0.90±0.27^{a}	0.78±0.25^{ab}	6.55±0.45^{ab}	7.16±0.57^{a}
Lock Roy	0.74±0.30^{a}	0.82±0.20^{a}	0.98±0.15^{a}	0.79±0.13^{ab}	7.44±0.92^{ab}	7.83±0.78^{ab}
Legend	1.34±0.52^{a}	1.60±0.63^{a}	0.96±0.12^{a}	0.93±0.13^{ab}	6.38±0.81^{ab}	7.14±0.80^{a}
Wall Brown	8.43±1.52^{c}	10.15±1.53^{c}	0.71±0.04^{a}	0.69±0.05^{ab}	6.48±0.44^{a}	6.90±0.42^{a}
Brown Spanish	11.21±1.19^{d}	13.18±1.95^{d}	2.56±0.37^{b}	2.37±0.27^{c}	10.93±0.48^{c}	11.33±0.46^{c}
Local Cultivar	98.90±1.14^{e}	99.44±0.47^{e}	3.06±0.68^{b}	3.16±0.29^{d}	6.90±1.07^{ab}	7.62±1.15^{a}
Mean	13.38±29.01	13.98±29.07	1.24±0.85	1.18±0.84	9.09±2.71	9.79±2.90

Value (Mean± SE) bearing common superscript (abcd) within column does not vary significant.

Table 8. Effect of set and seedling on dry matter, polar diameter and equatorial diameter of onion bulb

	Dry matter (%)		Polar diameter (cm)		Equatorial diameter (cm)	
Genotypes	Seedling	Set	Seedling	Set	Seedling	Set
Red cereole	14.71±0.76^{d}	15.50±0.72^{d}	4.80±0.68^{b}	5.03±0.53^{a}	6.12±0.31^{bc}	6.55±0.41^{bcd}
Katarina Red 3	14.18±0.82^{cd}	15.35±0.35^{d}	4.65±0.43^{ab}	4.78±0.40^{ab}	4.14±0.80^{ab}	4.55±0.49^{ab}
Katarina Red 7	13.59±1.22^{cd}	14.89±1.08^{d}	5.64±1.15^{bcd}	5.98±0.88^{bc}	5.02±0.31^{bc}	5.14±0.24^{abc}
Supreme	9.87±0.55^{b}	10.82±0.45^{b}	5.86±0.50^{bcd}	5.84±0.63^{bc}	6.21±0.33^{bc}	6.45±0.60^{cd}
Cyrus	8.20±0.20^{a}	8.83±0.51^{a}	6.62±0.99^{cd}	6.01±1.3^{b}	6.13±1.22^{c}	7.35±1.46^{d}
Lock Roy	9.09±0.75^{ab}	9.47±0.64^{ab}	5.47±0.55^{b}	5.80±0.49^{c}	6.59±0.87^{c}	6.91±0.62^{cd}
Legend	8.19±0.79^{ab}	8.48±1.04^{a}	6.74±0.74^{cd}	6.84±0.73^{c}	6.53±1.61^{c}	7.63±1.37^{d}
Wall Brown	8.21±0.38^{ab}	8.55±0.34^{a}	7.01±0.75^{d}	7.28±0.97^{c}	9.60±1.15^{d}	9.96±0.73^{c}
Brown Spanish	12.60±0.18^{c}	12.93±0.38^{c}	6.00±0.32^{bcd}	6.21±0.31^{bc}	5.78±0.81b^{c}	5.93±1.11^{bcd}
Local Cultivar	8.50±1.31^{ab}	9.37±1.03^{ab}	3.66±0.51^{a}	4.04±0.50^{a}	3.58±0.55^{a}	3.82±0.44^{a}
Mean	10.73±2.69	11.43±2.89	5.64±1.19	5.75±1.10	6.32±1.78	6.39±1.83

Value (Mean± SE) bearing common superscript (abcd) within column does not vary significant.

Table 9. Effect of set and seedling on average bulb weight and yield of onion bulb

	Average bulb weight (g)		Bulb yield (kg/ plot)	
Genotypes	Seedling	Set	Seedling	Set
Red Cereole	100.56±5.48^{abc}	107.37±9.30^{a}	11.62±1.22^{bc}	20.95±1.99^{a}
Katarina Red 3	81.73±7.28^{b}	91.37±5.51^{a}	9.20±1.44^{cd}	17.14±1.25^{a}
Katarina Red 7	92.89±6.94^{b}	107.32±6.76^{a}	11.40±1.28^{c}	20.94±1.21^{ab}
Supreme	114.63±17.88^{cd}	145.51±16.30^{b}	14.15±2.35^{c}	28.42±3.16^{c}
Cyrus	138.34±12.86^{ef}	185.04±11.24^{c}	14.47±1.16^{c}	34.15±1.23^{d}
Lock Roy	103.11±13.17^{bc}	145.57±13.59^{a}	12.56±1.93^{b}	28.51±2.40^{c}
Legend	138.36±4.29^{de}	194.12±10.12^{c}	15.10±0.60^{b}	37.77±2.24^{d}
Wall Brown	152.61±5.68^{f}	203.09±5.19^{c}	19.59±0.85^{c}	39.86±2.42^{d}
Brown Spanish	103.28±10.77^{c}	132.94±12.40^{b}	10.55±1.68^{ab}	25.22±2.52^{bc}
Local Cultivar	80.75±9.47^{a}	88.59±9.13^{a}	9.17±1.01^{ab}	16.73±1.81^{a}
Mean	109.53±24.55	140.09±41.74	12.73±3.17	26.96±8.09

Value (Mean± SE) bearing common superscript (abcd) within column does not vary significant.

The results indicated that for all the genotypes, planting through sets produced maximum effect on all the parameters compared to those grown through seedlings under agro-climatic conditions of district Leh, trans-Himalaya Ladakh, India. Genotype Wall Brown gave the best performance followed by Legend and Cyrus in terms of yield among all the genotypes under experiment.

has been reported by Kandil et al. (10). Similar results were obtained by Akhtar, (1), Gemma *et al*. (8). Soleymani and Shahrajabian (18) reported that foliage fresh weight, bulbing ratio, plant height, weight of bulb, total yield, favorite yield, total percentage of dry matter and nitrate content in bulb was significantly influenced by genotype. The stimulatory effect of sets on all the quantitative and qualitative traits could have been due to presence of more sprouting initials and reserved food material thereby resulting in better establishment and survival of plants under the adverse environmental conditions. Similar finding was reported by Khodadadi, (11) and Islam, (9).

3. Evaluation of different onion genotypes for mineral content as measured by ICP-OES at agroclimatic condition of Leh, trans-Himalaya, India

Materials and method

To determine the genetic variation of mineral accumulation, 10 genotypes of onion (*Allium cepa* L.) were selected, of which 9 had been obtained from BejoSheetal Seeds and one genotype (Local cultivar) from Leh. The seeds were sown for the production of seedlings in a trench. The research was conducted over two years; seeds were sown on 2nd March, 2013 in the first year and on 4th March, 2014 in the second year. Seedlings were planted on 6th May, 2013 and 7th May, 2014 in the experimental plots, in the first and second years, respectively.

The experiment was conducted in a randomized block design with four replications. The plot size was kept at 2.0 m^2 and the row-to-row and plant-to-plant spacings were 20 cm and 10 cm, respectively. All recommended agronomic practices were followed and plant protection measures were taken. In each year, the harvest was carried out in the first week of November. The onion bulbs were harvested (into standard 25 kg plastic nets and dried in ambient conditions for five weeks according to the standard practice in India. The dry aerial parts and roots were removed, and diseased or damaged bulbs were discarded.

The samples were taken randomly. Samples from each set of three bulbs from each genotype and each replication were combined and lyophilized prior to mineral analysis. The dry outer skins were removed, and a vertical wedge of tissue was taken from the basal section of the bulb. Each sample was weighed and immediately snap-frozen in liquid nitrogen, and stored at -40 ºC until it was lyophilized. Dry weight measurements were made on lyophilized samples.

1 g fine powder of onion bulb was taken in a 30 mL glazed porcelain crucible. The crucible was placed in a cool muffle furnace and muffled at 500 °C for 2 h. It was then removed from the

furnace, cooled and 3.0 mL HNO_3 was added to it. The sample was heated on a hot plate at 100 °C until it dried. The crucible was again placed in the muffle furnace, allowed to cool, and 10 mL HCl was added to it. The sample was transferred to a 50-ml volumetric flask, diluted to volume with deionized water and mixed thoroughly. All the elements were analyzed by using the Inductively Coupled Atomic Adsorption Spectrometer (Perkin-Elmer Optical Emission Spectrometer, Optima 7000 DV), 104 according to the methods in the Handbook of Reference Methods for Plant Analysis by Kalra (1998) and instruction given by supplier of ICP OES. In the experiment, each accession leaf sample was analyzed in triplicate and

12 elements were recorded (NO_2, NO_3, N, P, K, Ca, Mg, Na, Fe, Cu, Zn, Mn).

Genetic diversity was studied using the Mahalanobis (1936) generalized distance (D^2) extended by Rao (1952). Clustering of genotypes was done according to Tocher's Method. The average intra-cluster distance was calculated by the following formula as suggested by Singh and Choudhary (1985). Trait variability analysis was performed by the principal component analysis (PCA) method, with the number of principal components being chosen based on the screen test (Kovacic, 1994). Agglomerative Hierarchical cluster analysis was used to determine differences and similarities among the genotypes, and the distance measure used was Gower General Similarity Coefficient as the parameter that best reflects the differences existing among the genotypes (Kendall, 1980). All statistical analyses were carried out based on 10 mineral element using SPSS 21.

Results

The evaluation of mineral content provided significant information that could be used for the genetic advancement of onion. A total of 10 mineral elements and 10 onion genotypes were investigated, and considerable variation was observed in most of the concentrations. All the genotypes were grouped into three different clusters (table 10). Genotypes grouped into Cluster

II and III showed the maximum inter-cluster diversity (table 11). From cluster mean values, genotypes in Cluster I, II and III deserve consideration for their direct use as parents in hybridization programs to develop high-mineral content in long-day onion varieties. There is significant genetic variability among the tested genotypes that indicates the presence of an excellent opportunity to bring about improvement through wide hybridization by crossing genotypes in different clusters. Further studies on the influence of environment and agronomic practices on the genetic potential of the varieties in different long-day onion environments are necessary. This is helpful in the stratification of the environments, based on the mineral content and stability. From the results, it can be concluded that genotypes from cluster I for Mg, K and B (table 12), genotypes for Ca, P, Na, Zn and Cu from Cluster II and genotypes belong to cluster III for Fe and Mn could be selected as parents for hybridization programme. Crosses involving parents belonging to more divergent clusters would be expected to manifest maximum heterosis and a wide variability in genetic architecture.

Table 10.Distribution of ten genotypes of onion in different clusters

Sr.no.	**Cluster**	**Genotypes**
1	I	Katrina Red 7, Katrina Red-3, Supreme, Cyrus
2	II	Lock Roy, Legend, Wall Brown, Brown Spanish, Local Cultivar
3	III	Red Cereole

Table 11. Average intra- (bold face) and inter-cluster distance (D2) of 10 onion genotypes

Sr. no.	**Clusters**	**I**	**II**	**III**
1	I	**2.050**		
2	II	3.892	**2 .322**	
3	III	4.352	4.371	**0.000**

Table 12. Mean values for three clusters based on ten mineral elements

Sr. no.	**Minerals**	**Clusters**		
		I	II	III
1	Ca	101.36	116.48	38.21
2	Fe	1.52	2.59	3.21
3	Mg	100.19	88.42	91.17
4	P	92.02	151.73	148.60
5	K	212.70	197.90	122.40
6	Na	30.91	49.60	31.47
7	Zn	1.91	3.17	1.70
8	Mn	1.42	1.56	1.70

9	Cu	0.63	0.71	0.55
10	B	0.91	0.42	0.67

4. Evaluation of onion genotypes for post-harvest keeping quality of bulb at agro climatic condition of Ladakh.

Material and methods

The present study was conducted during two consecutive cropping seasons 2013 and 2014 at vegetable research unit of Defence Institute of High Altitude Research.The seed was sown into trench with 5cm spacing between genotypes during the first week of April. At that time all standard agronomic practices recommended for onion was done. After 60 days, when seedling obtained optimum set/baby bulb size, it was harvested and cured into the shade for one month. It was stored at 2±1 ^{0}C and 75±2 % relative humidity in onion store.

The experimental material consisted of 9 long day genotypes and one local cultivar of onion viz. Red Cereole, Katarina Red 3, Katarina Red 7, Supreme, Cyrus, Lock Roy, Legend, Wall Brown, Brown Spanish and a Local Cultivar, respectively. Sets were planted on 18 May 2013 to produce mature onion bulbs using standard agronomic practices.

The crop was harvested at maturity when 70 per cent of the plants showed drying and falling of their tops. The plants were pulled along with leaves and kept for three days in the field for curing. The dry aerial parts were removed with sharp clean knives leaving 2.5 cm top above the bulb. These bulbs were kept under 50 per cent shade for curing for 20 days. The cured onion bulbs were sorted out; any diseased or damaged bulbs were discarded prior to storage and five kg healthy bulbs from each treatment were packed in thin gunny bag and kept in laboratory for storage studies. Bulbs were stored under controlled atmosphere (CA) conditions 2±1ºC and 75±2% Relative humidity.

Experimental design

The experiment was conducted in randomized block design with three replications. Onion bulbs of long day genotypes viz. Red Cereole, Katarina Red 3, Katarina Red 7, Supreme, Cyrus, Lock Roy, Legend, Wall Brown, Brown Spanish and a Local Cultivar were stored for 50, 100, 150 days in controlled atmosphere2±1ºC and 75±1% relative humidity. The observations viz. dry matter, TSS, hardiness of bulb, total sugar, reducing sugar and non-reducing sugar, loss in weight (%), Rotting (%) Sprouting (%), Sprout length (cm), Incident of black mold (%), Marketable bulbs (%) were recorded throughout the storage.

Dry matter (%)

Bulbs were randomly selected from each treatment and cut into small pieces with the help of stainless steel knife. A known weight of the sample was dried in hot air oven at 60°C temperature till a constant weight was obtained. The per cent dry matter was calculated by the following formula.

Total Soluble Solids (%)

The total soluble solids (TSS) of onion were determined using a hand refractometer (Attago, Japan.) The values were expressed as per cent total soluble solids of the bulbs according to AOAC (2005).

Hardness of the bulb (kg/cm^2)

The hardness of onion bulbs was measured using a hand penetrometer (Fruit Pressure Tester, Make: Effegi, Model: PT 327) and the pressure required to penetrate the bulb was recorded in kg per cm2.

Ascorbic Acid (mg/100 g)

The vitamin C (Ascorbic acid) in onion bulb was calculated by titrimetric method as described by AOAC (2005).

Total Sugars (%)

Total Sugar was estimated by Dubois et al., (1950) Method.

Reducing sugar (%)

Reducing sugar in the bulb was determined as per the Dinitrosalicylic acid method (Miller, 1972). The values obtained were expressed as percentage on fresh weight basis.

Non-reducing sugar (%)

The percentage of non-reducing sugar was obtained by subtracting the values of reducing sugar from that of total sugar and multiplying the same with 0.95 as given below (Somogyi, 1952).

Non-reducing sugar (%) = (Total sugar - reducing sugar) x 0.95

Weight Loss (%)

The weight of the bulbs was using an electronic balance. The cumulative loss in weight of bulbs was calculated and expressed as per cent weight loss.

Rotting percentage

The weight of the rotted bulbs at the end of 50, 100 and 150 DOS was recorded under each storage condition and the rotting percentage was calculated by using the formula

Sprouting percentage

For determining the sprouting percentage on stipulated days after storage, the bulbs showing a sprout were separated from the lot and weighed on an electronic balance. The sprouting percentage, which indicated the weight of the bulbs sprouted on 50, 100 and 150 DOS was calculated.

Sprout length (cm)

Five sprouted bulbs were randomly selected and length of the sprout in each bulb was measured. The mean length of sprout was expressed in centimeters.

Incidence of black mold (%)

The incidence of black mould was expressed as percentage of bulbs affected out of 100 bulbs.

Marketable bulbs (%)

At the end of storage period (150DAS), the rotted and sprouted bulbs were separated and the weight of healthy bulbs was recorded. The recovery of marketable bulbs was calculated by using the following formula.

Marketable bulbs (%) =Weight of the healthy bulbs obtained/Initial weight of bulbs stored×100

Statistical Analysis

All statistical analyses in this research were carried out using statistical package "SPSS" for Windows Version 21. Least significant difference between mean values was calculated using Duncan's Multiple Range Test (DMRT) at 5% significance level.

Result

For all the genotypes, Dry matter, TSS, Total sugar and Non-reducing sugars, rotting (%) Sprouting (%), Sprout length (cm), Incident of black mold increased gradually during storage. The greatest increase is observed during last 150 days of storage. While hardiness of bulb, ascorbic acid, reducing sugar, physiological loss in weight, marketable bulbs was decreased gradually during storage period. The greatest decrease was observed during the 150 days of storage. For storage period, similar pattern of increase and decrease of all the observed traits were shown by all the genotypes. On the other hand at genotypic level, significant variation was observed for their storage potential. Genotypes Red cereole, Katarina red 3 and Katarina red 7 gave highest marketable bulb at the end of storage.

It is hypothesized that the storage potential of bulbs of different onion genotypes were negatively related to the time. Onion genotypes Red cereole, Katarina red 3 and Katarina red 7 were characterized by having good storage potential that could be stored overwinter at high altitude and maintain the supply overwinter. Meanwhile, this could help to extend the onion bulb availability period in temperate regions.

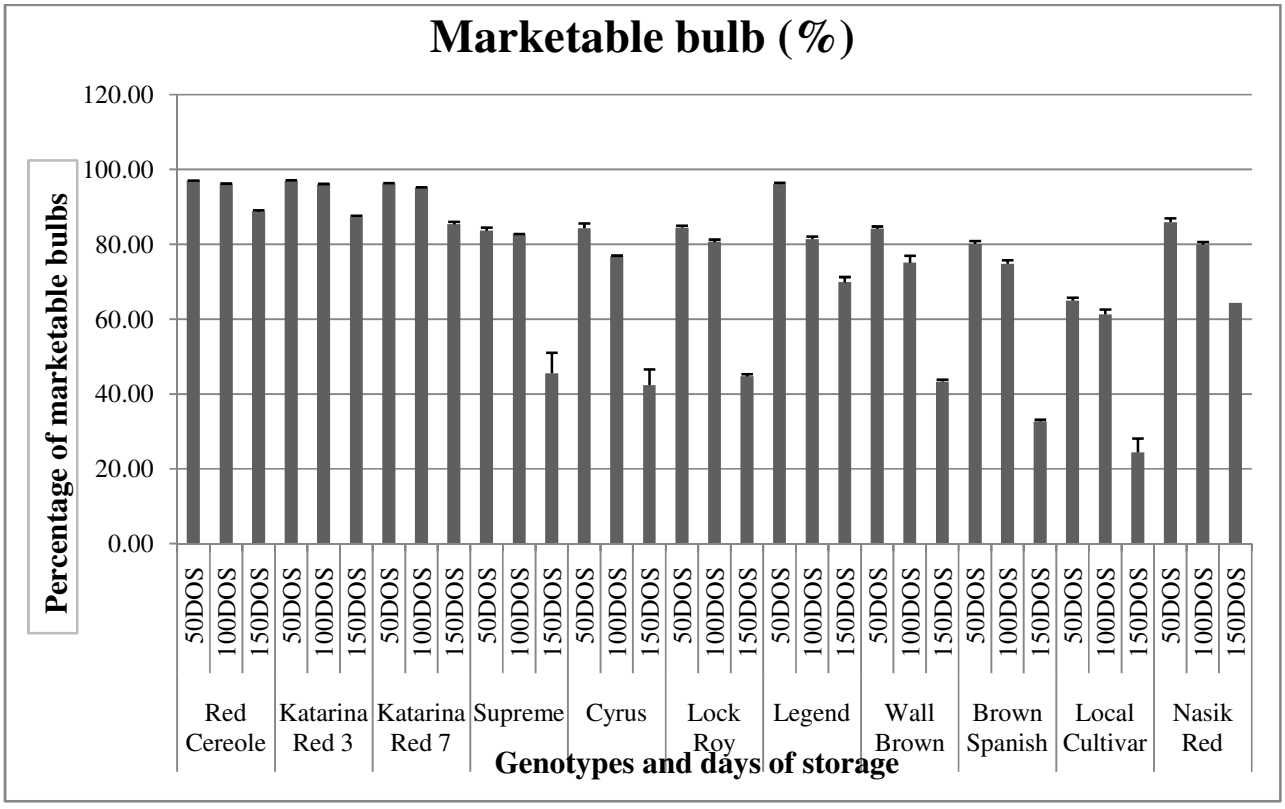

Fig. 4 shows the percentage of Marketable bulb of studied onion genotypes after different storage durations

It might be due to the genetic potential of these genotypes, which produces high amount of TSS, and Dry matter content that minimizes to lack of weight loss, sprouting percent, incidence of black mold (Rafika et al., 2006.

Conclusion

1. Genotypes belong to cluster I, II and IV can be directly used as parent in hybridization program. To develop high yielding onion hybrids and to improve existing cultivar.
2. Amongst the genotypes, Wall Brown followed by Legend and Cyrus gave highest bulb yield raised through sets.
3. Genotypes belong to cluster II and III can be directly used as parent in crop improvement program. To develop mineral rich onion hybrids and to improve the existing onion cultivars.
4. Onion genotypes Red cereole, Katarina red 3 and Katarina red 7 were characterised by having good storage potential that could be stored overwinter at high altitude and maintain the supply overwinter.

References

1. Akhtar M E, Khurran B, Khan M Z, Khakhar K M (2002). Effect of potash application on yield of different varieties of onion (*Allium cepa* L.). Asian Journal of Plant Sciences 1 (4): 324-325.
2. Gemma A C, Terry L A, White PJ (2007). Effect of controlled atmosphere storage on abscisic acid concentration and other biochemical attribute of onion bulbs. Post-harvest Biology and Technology 39:233-242.
3. Kandil A A, Leilah A A, Mostafa A K, Fathalla F H(2010). Study on the internal bulb quality of some new Egyptian onion cultivars under different irrigation regimes. Journal of Plant Production 1 (2): 205-212.
4. Soleymani, A. and Shahrajabian, M. H. 2012. Effects of different levels of nitrogen on yield and nitrate content of four spring onion genotypes. Inter. J. of Agric. and Crop Sci., 4(4): 179-182.
5. Khodadadi M(2012). The effect of planting date and motheral bulb size on quantitative and qualitative seed traits of onion red rey variety. International Jornal of Agriculture 2(4):324-327.
6. Islam K S(2002). Control of growth cycle of onion for high yield and quality seed production. Ph.D. Thesis, Department of Horticulture, Bangladesh Agricultural University, Mymensingh, Bangladesh.
7. Mahalanobis PC (1936). On the generalized distances in statistics. Proceedings of National Institute of Sciences (India) 2:49-55.
8. Kalra YP (1998). Handbook of Reference Method for Plant Analysis. CRC Press, Taylor & Francis group, Boca Raton, FL. classification. WH Freman and Company, San Francisco.
9. Singh RK, Choudhary BD (1985). Diametrical Methods in Quantitative Genetic Analysis. Kalyani Publishers, New Delhi.

10. Rao CR (1952). Advanced Statistical Methods in Biometrics Research John Wiley and Sons, New York.
11. Kendall M (1980). Multivariate Analysis. 2nd ed. Charles Griffin and Co London.
12. Kovacic, Z. (1994): Multivariate analysis (in Serbian). University of Belgrade, Faculty of Economics, pp.282.
13. Selvaraj S (1976) Onion: Queen of the kitchen. KisanWorld, 3(12): 32-34.
14. Augusti KT (1990) Therapeutic and medicinal values of onion and garlic. In Onion and Allied Crops. Ed. Brewster, J.L. and Rabinowith, H.D., BacoRatani, Florida, CRC Press, 3: 93-108.
15. SKUAST-K, Leh (1998). KVK Leh Annual Progress Report (1997-98 and Action Plant 1998-99), DEE, SKUAST, RARS, Leh, pp 32.
16. Azmat M A, Nawab N N, Khan A A, Ashraf M, Niaz S, Mahmood K (2011). Characterization of pea germplasm International Journal of Vegetable Science 17: 246–258.
17. Singh SK, Singh RS, Maurya DM, Verma OP (1987). Genetic divergence among lowland rice cultivars: Annual report of indian agricultural research institute (IARI), New Delhi, India.
18. Akter MS, Biswas A, Siddique SS, Hossain S, Ivy NA (2015). Estimation of genetic diversity in Onion (*Allium cepa*L.).The Agriculturists. 13(1):26-34.
19. Ghaderi A, Adams MW, Nassib AM (1984). Relationship between genetic distance and heterosis for yield and morphological traits in dry edible bean and faba bean. Crop Sciences. 24:37-42.
20. Kandil AA, Leilah AA, Mostafa AK, Fathalla FH (2010). Study on the internal bulb quality of some new Egyptian onion cultivars under different irrigation regimes. International Journal of Plant Prod. 1(2):205-212.

List of Publications

1. **Arya JS**, Narendra Singh, Preeti Singh Arya and Anil Kant (2017) Comparative performance of onion genotypes using 'Sets and Seedlings' as planting material at Leh cold desert. Journal of Applied Horticulture, 20(1): 64-68.

2. **Arya JS** , Narendra Singh, Preeti Singh Arya and Anil Kant (2017)Morphological variations and relationship among onion germplasm for quantitative and qualitative traits at trans-Himalaya Ladakh, India. Australian Journal of Crop Science 11(03):329-337.

3. **Arya JS**, Singh N, Rinchen T, MauryaSB and Korekar G (2015): Genotypes, geographical regions and solvents dependent antioxidant activity of *Rumex patientia* L. in cold desert of trans-Himalaya Ladakh, India. Australian Journal of Crop Science.9 (2):98-104.

4. **Arya JS**, Preeti, Kumar M and Chand P (2014) Analysis of genetic divergence for yield Contributing traits in wheat (*Triticum aestivum* L.) Genotypes. Progressive Agriculture. 14(1): 178-184.

5. Singh N, Maurya SB, **AryaJS** and Srivastava RB (2013) 'rootlet to seed': a new technology for high quality carrot seed production in one year. Current science 104(9):1132-33.

6. Singh N, **Arya JS**, Maurya SB and Srivastava RB (2013): Rumex (*Rumex patientia* L.) Spinach of high altitude cold desert. Current Science 104(5):574.

7. **J.S. Arya,** Pooran Chand, S.K. Singh and Gaurav Kumar (2011) Correlation and Path analysis in bread wheat (*Triticum aestivum* L.) International conference on Issues for climate change, Land use diversification and biotechnological tools for livelihood security, October 8-10.

8. Gaurav Kumar, S.K. Singh, Pooran Chand, B. Singh and **J.S. Arya** (2011) Character association and path analysis inwheat (*Triticum aestivum* L.) International conference on Issues for climate change, Land use diversification and biotechnological tools for livelihood security, October 8-10.

Conference Presentation

1. Jagdish Singh Arya, Preeti, Narendra Singh and Kalyani Srinivasan (2016) Effect on Yield and Quality Attributes of Onion Grown Through Set and Seedling under Agro-Climatic Conditions of District Leh, Trans-Himalaya Ladakh, India. 7th Indian Horticulture Congress, November 15-18, 2016, New Delhi.

2. Jagdish Singh Arya and Narendra Singh (2014) Morphological Variations and Relationship among Onion Germplasm for Quantitative and Qualitative Traits at trans-Himalaya Ladakh, India, 6th Indian Horticulture Congress, November 6-9, 2014, CODISSIA Trade Fair Complex Coimbatore.